

U.S.S.N. 10/607,903

Filed: June 27, 2003

AMENDMENT AND RESPONSE TO OFFICE ACTION

Amendment

1. (currently amended) A bacterial strain ~~producing~~ for production of a fermentation product to be isolated, wherein the product is selected from the group consisting of antibiotics, organic acids, amino acids, proteins, vitamins, polyhydroxyalkanoates, and polysaccharides, and wherein the bacteria has been bacterial strain is genetically modified to express a heterologous nuclease gene ~~or a genetically modified homologous nuclease gene or mutated to improve the activity of a homologous or heterologous nuclease gene~~, wherein the nuclease ~~protein gene product~~ is secreted into the periplasmic space or culture medium in an amount effective to degrade at least 95% of all of the nucleic acid released following lysis of the cells in less than 24 hours and reduce the viscosity of a cell lysate in a bacterial cell culture having a density of at least 50 g/l so that recovery of the product is enhanced.
2. (currently amended) The ~~baetaria~~ bacterial strain of claim 1 which is capable of growth to cell densities of at least 50g/l.
3. (original) The bacterial strain of claim 2 which produces a polyhydroxyalkanoate to levels of at least 40% of its dry cell weight.
4. (currently amended) The bacterial strain of claim ~~24~~ 1 for use in an aqueous process to manufacture poly(3-hydroxyalkanoates) granule suspension which is essentially free of nucleic acids.

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5. (original) The bacterial strain of claim 1 for use in a process for making polysaccharides selected from the group consisting of xanthan gum, alginates, gellan gum, zooglan, hyaluronic acid, and microbial cellulose.
6. (original) The bacterial strain of claim 1 wherein the nuclease gene is a heterologous gene obtained from an organism other than the bacterial strain.
7. (original) The bacterial strain of claim 1 in which the nuclease gene is integrated into a host strain selected from the group consisting of *Ralstonia eutropha*, *Methylobacterium organophilum*, *Methylobacterium extorquens*, *Aeromonas caviae*, *Azotobacter vinelandii*, *Alcaligenes latus*, *Pseudomonas oleovorans*, *Pseudomonas fluorescens*, *Pseudomonas putida*, *Pseudomonas aeruginosa*, *Pseudomonas acidophila*, *Pseudomonas resinovorans*, *Escherichia coli*, and *Klebsiella*.
8. (original) The bacterial strain of claim 1 wherein the nuclease is expressed in an amount effective to degrade at least 95% of all of the nucleic acid released following lysis of the cells in less than 24 hours.
9. (currently amended) The bacterial strain of claim 1 wherein the nuclease is encoded by a homologous nuclease gene which has been modified to enhance nuclease activity.
10. (original) The bacterial strain of claim 9 wherein the bacterial strain is mutated and the mutated bacteria are screened for an increased amount of nuclease activity.
11. (currently amended) A fermentation process comprising adding to a growth medium a bacterial strain ~~producing~~ for production of a fermentation product to be isolated, wherein the product is selected from the group consisting of antibiotics,

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organic acids, amino acids, proteins, vitamins, polyhydroxyalkanoates, and polysaccharides, and wherein the ~~bacteria has been~~ bacterial strain is genetically modified to express a heterologous nuclease gene ~~or a genetically modified homologous nuclease gene~~ or mutated to improve the activity of a homologous or heterologous nuclease gene, wherein the nuclease ~~protein gene~~ product is secreted into the periplasmic space or culture medium in an amount effective to degrade at least 95% of all of the nucleic acid released following lysis of the cells in less than 24 hours and reduce the viscosity of a cell lysate in a bacterial cell culture having a density of at least 50 g/l so that recovery of the product is enhanced.

12. (original) The method of claim 11 wherein the bacterial strain is grown to cell densities of at least 50 g/l.

13. (currently amended) The ~~method~~ bacterial strain of claim ~~11~~ 1 wherein the bacterial strain produces a polyhydroxyalkanoate.

14. (original) The method of claim 13 further comprising growing the bacterial strain to produce levels of at least 40% of its dry cell weight.

15. (original) The method of claim 11 further comprising lysing the cells.

16. (original) The method of claim 14 further comprising using an aqueous process to manufacture a poly(3-hydroxyalkanoates) granule suspension which is essentially free of nucleic acids.

17. (original) The method of claim 11 used in a process for making polysaccharides selected from the group consisting of xanthan gum, alginates, gellan gum, zooglan, hyaluronic acid, and microbial cellulose.

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18. (currently amended) The ~~method~~ bacterial strain of claim ~~11~~ 1 wherein the fermentation product is a protein and the protein is used in a process for making intracellular proteins selected from the group consisting of enzymes, growth factors, and cytokines.
19. (original) The method of claim 11 wherein the nuclease gene is integrated into a host strain selected from the group consisting of *Ralstonia eutropha*, *Methylobacterium organophilum*, *Methylobacterium extorquens*, *Aeromonas caviae*, *Azotobacter vinelandii*, *Alcaligenes latus*, *Pseudomonas oleovorans*, *Pseudomonas fluorescens*, *Pseudomonas putida*, *Pseudomonas aeruginosa*, *Pseudomonas acidophila*, *Pseudomonas resinovorans*, *Escherichia coli*, and *Klebsiella*.
20. (original) The method of claim 11 wherein the strain secretes nuclease into the periplasm or growth medium.
21. (original) The method of claim 11 wherein the strain expresses nuclease into the growth medium in an amount effective to degrade at least 95% of all of the nucleic acid released following lysis of cells in the growth medium in less than 24 hours.
22. (original) The method of claim 11 wherein the bacterial strain expresses a homologous nuclease, further comprising mutating the bacterial strain and screening for bacteria expressing enhanced nuclease activity.
23. (original) The method of claim 11 wherein the bacteria expresses a homologous nuclease, further comprising genetically engineering the nuclease to enhance nuclease activity.